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ABSTRACT

A study of Kagan's peer play situation for measuring heritable influences on children's behavioral inhibition (that is, shyness) drew its data from the large, multimethod, multivariate MacArthur Longitudinal Twin Study. The MacArthur study examined behavioral inhibition across four ages and two situations, looking for possible sex differences and the heritability of behavioral inhibition at each age. Subjects for the project were approximately 100 pairs of same-sex twins who were tested longitudinally at 14, 20, 24, and 36 months. At each age, children were placed in various situations: in the presence of a stranger, an intimidating object, or peers; and with or without their twin. Children's reactions, and the amount of time children spent near their mothers, were measured. Stability of behavioral inhibition was observed across ages in the same situations, and to a lesser extent across both situations and ages. No sex differences were found. Measurements of the peer play situation seemed to demonstrate an aspect of inhibition with a highly heritable component, especially among extremely inhibited children. (SAK)

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Longitudinal and Behavior Genetic Analyses of Inhibition From 14 to 36 Months: The MacArthur Longitudinal Twin Study

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Behavioral Inhibition is defined by a specific set of responses to strange or unfamiliar situations. In particular, it involves becoming quiet, ceasing activity, and withdrawing or retreating from the people or places that are unfamiliar.

Kagan and Reznick and their colleagues (e.g., Kagan, Reznick, Clarke, Snidman, & Garcia-Coll, 1984; Kagan, Reznick, & Gibbons, 1989; Kagan, Reznick, Snidman, Gibbons, & Johnson, 1988) have provided convincing evidence for the longitudinal stability of behavioral inhibition. Through measures such as the peer play situation, assessment of psychophysical measures, and laboratory testing of the children in unusual situations, they have demonstrated that extremely inhibited children tend to preserve that attribute from infancy through the early school years. In particular, behaviors such as maintaining proximity to mother, amount of interaction with unfamiliar people, and cessation of play activity are representative of those children who are rated as extremely inhibited.

Behavior genetic studies have shown that there appears to be a strong genetic component for behavioral inhibition across the lifespan. Methods of assessing shyness in infancy and childhood have ranged from parental questionnaires to laboratory ratings of fearfulness and emotional tone. Matheny (1988), for example, found significantly larger MZ correlations than DZ correlations on three measures of inhibition from 18 to 30 months. Kagan's peer play situation, however, has not yet been assessed for heritable influences. My talk today will address this.

The project that my data are drawn from is the MacArthur Longitudinal Twin Study. It is an exciting project because eight investigators from six different universities collaborate on it, and each brings his or her expertise

in different areas, which results in a large, multimethod, multivariate study intending to address the relationships between a large number of social and cognitive variables in young childhood. The behavioral inhibition variables which I will focus on in this talk comprise one aspect of this large project. The other two people with whom I am working closely on this project and whom I wish to acknowledge are Jerome Kagan and Steven Reznick.

The subjects in this project are approximately 100 pairs of same-sex twins tested longitudinally at 14, 20, 24, and 36 months. All twins are selected to be within the normal range for birthweight and gestational age at birth, and they must have survived without extended hospitalization or evidence of abnormality.

I had three main goals for this study: to examine behavioral inhibition across 4 ages and 2 situations; to look for any possible sex differences; and to examine the heritability of behavioral inhibition at each age.

At each age the twins come to the laboratory, which is in Boulder, CO, and are administered a variety of measures. At 14 and 20 months, one of these involves one of the twins at a time playing in a room with his or her mother and then having a stranger enter the room and being presented with a strange and somewhat intimidating object. The child's reactions to the unfamiliar person and object are rated and two indexes are formed. The first (Index 1) is based on the amount of time which the child spends proximal to mother, whether or not the child retreats from the stranger or the object, and whether or not the child ever approaches the stranger or the object. The second index (Index 2) is simply the mean time the child spends proximal to mother.

At 24 and 36 months, the twins are placed in a peer play situation. At 24 months, two pairs of twins - four children - and their mothers are brought

into a laboratory playroom and the children are allowed to play freely for 25 minutes. During this time, four trained coders located behind one-way mirrors code the children; each coder is assigned to a particular child. The children are coded on 10 behaviors which were chosen because they were thought to be related to inhibited behavior.

At 36 months, the twins are separated during the peer play situation, so one child from each pair and the two mothers enter a laboratory playroom and the children are allowed to play for 25 minutes. After this time, the other two children, one from each pair, and their mothers are brought into the playroom and again the two children are allowed to play for 25 minutes. The children's behaviors again are coded on the same 10 variables by two coders situated behind a one-way mirror.

Three variables consistently have shown high inter-correlations and face validity at each age. These three behaviors were: Time spent proximal to mother; Time spent staring at one of the unfamiliar twins; and Number of approaches made toward one of the unfamiliar twins (inverse). These behaviors were normed and averaged to form an Inhibition Aggregate score at each age.

The first aim of this study was to examine the relationships of the inhibition variables across ages and situations. Pearson correlations are presented in Tables 1 and 2. We can see from Table 1 that the inhibition indexes at 14 and 20 months are significantly correlated across the two ages at about .4. Similarly, as seen in Table 2, the peer play Aggregate is significantly correlated across the two ages, $\underline{r} = .35$. The correlations are, not surprisingly, less strong between the different measures across ages, but it can be seen that Index 2 at 20 months is significantly correlated with the peer play Aggregate at both 24 and 36 months.

Insert Tables 1 and 2 about here

I will just mention here that the sex difference analyses showed virtually no sex differences for the inhibition variables at any of the ages.

The third aim was to examine genetic, and in turn environmental, effects for each variable at each age. The first step was to compute MZ and DZ correlations to get a rough estimate of heritability. Briefly, MZ twins share their genetic makeup entirely, whereas DZ twins share on average 50% of their genes, so a comparison between the two correlations can give us an idea of the extent to which a given trait is heritable. (For a more complete description of this methodology, see e.g., Plomin, DeFries, and McClearn, 1990.) Behavior genetic results for this talk will concentrate on the peer play Inhibition Aggregate.

As can be seen in Table 3, at 24 months the heritability estimate is huge, decreasing to a more reasonable estimate at 36 months. The estimate at 24 months is really much too large and may be a result of nonadditive genetic effects, which only MZ twins share, or it may be a function of the testing situation. Remember that at 24 months the twins play in the room together. You can see by the correlations that even the DZ twins are much more similar to each other than are any of the twins at 36 months, and this also is true of the correlations at 14 and 20 months. This effect is even more pronounced for the MZ twins. It appears that being in the room together fosters an increase in behavioral similarity, so that the genetic influence that exists is being magnified. At 36 months, when the twins are once again separated, the heritability drops back to a more reasonable estimate which is consistent with

the estimate of .20 found at 20 months.

Insert Table 3 about here

More sophisticated analyses are needed to more completely explore the genetic and environmental etiology of behavioral inhibition. The rationale for the following multiple regression approach is summarized in Figure 1. First, the top curve shows the general population rated on a particular trait, in this case behavioral inhibition. Those children who are rated as extremely inhibited are represented by the hatched area at the right extreme of the curve. These children are called the "probands". The idea is that if there is a genetic component to behavioral inhibition, then MZ cotwins will be more similar to their extremely inhibited proband twins than DZ cotwins will be to their proband twins. Thus, we would expect to see a greater regression toward the mean for the DZ cotwins than for the MZ cotwins. The second curve in Figure 1 illustrates the normal curve for the MZ cotwins of extremely inhibited proband twins. You can see that their mean, $\bar{C}_{MZ}$, is shifted to the left of the proband mean, $\bar{P}$, but is shifted markedly to the right of the population mean, μ. The third curve is for the DZ cotwins of the extremely inhibited probands. Their mean, $\bar{C}_{DZ}$, is shifted even farther to the left of the proband mean, $\bar{P}$. Thus, simplistically, if $\bar{C}_{MZ}$ is significantly greater than $\bar{C}_{DZ}$, then we can assume that there is an important effect of genetic variability on the trait in question, in this case behavioral inhibition.

Insert Figure 1 about here

The multiple regression approach which was used has been formulated by DeFries and Fulker (1985, 1988). This analysis was undertaken for the 24 month peer play data. Only the extremely inhibited children were included in this analysis to determine whether extreme inhibition has a genetic component. If so, then the MZ cotwins, that is, the twins of the extremely inhibited MZ twins, will be rated as more inhibited than the DZ cotwins. The 52 children in the top 20% of the distribution and their cotwins were included in this analysis.

The proband means were transformed by dividing each proband score by the proband mean within each zygosity. The cotwin scores also were divided by the proband mean. The population mean was zero because of the way the Inhibition Aggregate was formed. You can see in Table 4 that the MZ cotwins have a significantly higher mean inhibition score, .51, than do that DZ cotwins .01.

Insert Table 4 about here

The probandwise concordance rates also are presented in Table 4 to show you the different concordance rates for MZs and DZs for the extremely inhibited twins. Clearly, the MZs were much more concordant for behavioral inhibition than were the DZs.

Beta$_2$ provides an index of the extent to which the deviant scores of the probands are due to heritable influences. You can see that it was very high. 1.0 should be the upper limit for a heritability estimate, and B_2 has reached its ceiling. This suggests that behavioral inhibition as measured by the peer play is highly heritable. A caveat is that we are dealing with a small sample here, only 52 pairs, because this is an analysis of the extreme scorers. A

larger sample would yield a more reliable heritability estimate.

The bottom line here is that, first, stability of behavioral inhibition has been observed across ages within the same situation and to a lesser extent across both situations and ages. Secondly, the peer play situation seems to measure an aspect of inhibition that has a highly heritable component, especially when we examine the extremely inhibited children. Keep in mind that at 24 months the twins are in the situation together. Further analysis of the effects of sibling interaction on the heritability estimate can be made by comparing the inhibition behaviors of the children at 24 months with those at 36 months when the children are separated during this procedure. I am working on those analyses now. They should provide us with further insight into the shared environmental effects important for the display of inhibited behavior.

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Table 1

CORRELATIONS OF INHIBITION AGGREGATE MEASURES

	14 MONTHS		20 MONTHS	
	INDEX 1	INDEX 2	INDEX 1	INDEX 2
INDEX 1 14 MO.	1.00	.93***	.43***	.42***
INDEX 2 14 MO.		1.00	.44***	.45***
INDEX 1 20 MO.			1.00	.95***
INDEX 2 20 MO.				1.00

Table 2

CORRELATIONS OF INHIBITION PEER PLAY MEASURES

(Ns in parantheses)

	24 MONTHS	36 MONTHS
INDEX 1 14 MO.	.07 (101)	.16 (81)
INDEX 2 14 MO.	.10 (106)	.21* (81)
INDEX 1 20 MO.	.22* (83)	.18 (75)
INDEX 2 20 MO.	.30** (83)	.23* (75)
PEER PLAY 24 MO.	1.00	.35*** (85)

Table 3

CORRELATIONS AND HERITABILITY ESTIMATES

(Ns in parantheses)

	MZ	DZ	h^2
PEER PLAY 24 MO.	.86 (74)	.44 (58)	.84
PEER PLAY 36 MO.	.28 (51)	.00 (31)	.28

Table 4

EXTREMELY INHIBITED CHILDREN -

24 MONTH PEER PLAY AGGREGATE

ZYGOSITY	PROBAND MEAN	COTWIN MEAN	PROBANDWISE CONCORDANCE RATE	B_2
AFTER TRANSFORMATION				
MZ	1.00	.51	.49	1.02
DZ	1.00	.01	.13	

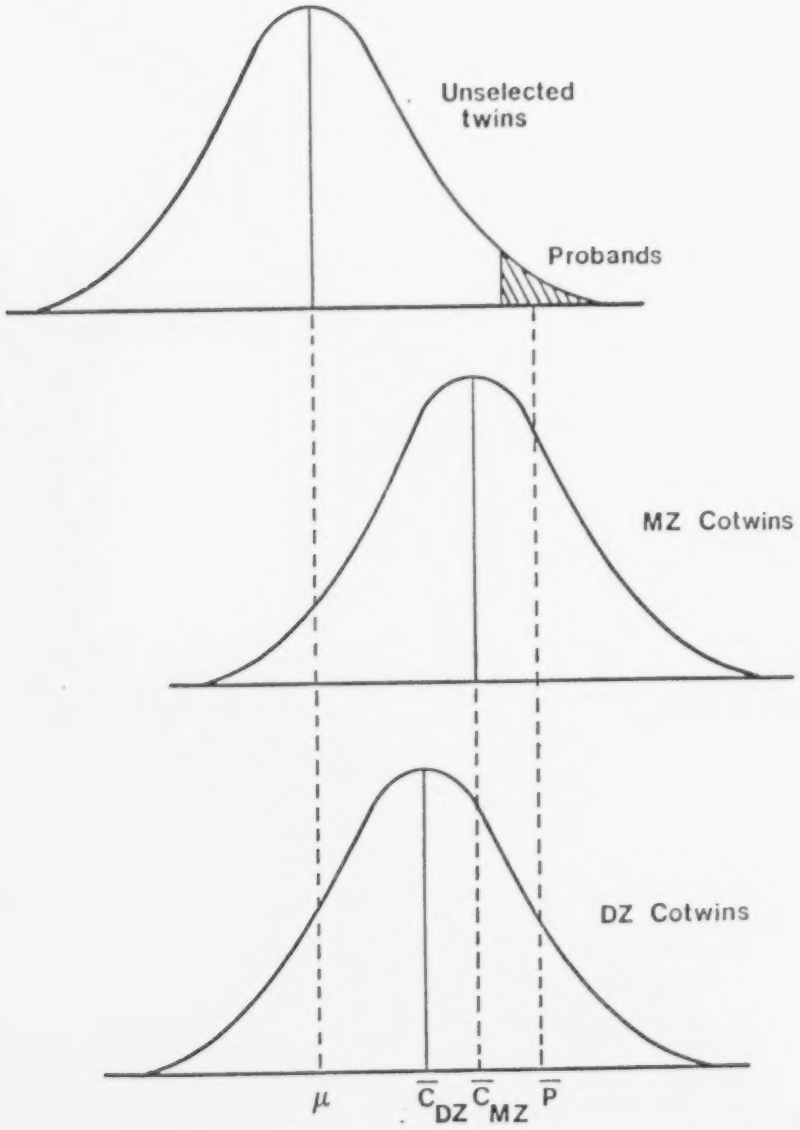
Unselected twins
Probands
MZ Cotwins
DZ Cotwins
μ
$\overline{C}_{DZ}$
$\overline{C}_{MZ}$
$\overline{P}$

Figure 1